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METHODS AND COMPOSITIONS FOR PRODUCTION OF RECOMBINANT PEPTIDES

FIELD OF THE INVENTION

This invention entails a method for solubilizing and recovering, in bioactive and isolated form with retained native state configuration, target peptide from a host organism in which the heterologous polypeptide is present in insoluble form. Broadly this method comprises (i) disrupting the host cell to produce a lysate (ii) recovering lysate precipitate containing the polypeptide (iii) resuspending the lysate precipitate in a denaturant-free, non-buffered solubilization solution to produce a solubilization preparation that optimally comprises sodium hydroxide between about 8 and about 10 mM, Mannitol between about 2 and about 2.5 mM, Lactose between about 1 and about 2 mM and the target peptide between about 1 and about 4 mg peptide per ml solubilization solution, wherein the resultant solubilization preparation has a pH of between about 9 and about 11.2; (iv) recovering supernatant from the solubilization preparation containing non-denatured target peptide. The invention further comprises isolated insoluble proteins in bioactive form and native state configuration.

BACKGROUND OF THE INVENTION

Many peptides, polypeptides, and proteins (collectively, "target peptide(s)") can be produced via recombinant means. Recombinant protein production has been established in a variety of expression systems. Such expression systems, include strains of bacteria and fungi as well as mammalian and baculovirus or insect cells. These expression systems are not without technical problems. One problem is the recovery or separation of the target peptide from the system as a whole.

Isolating a target peptide from native or host cell/expression system proteins and other cellular products is a significant hurdle in expression system utility. Consider, for example, yeast systems employed for synthesis of target peptides such as human

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